

<http://dx.doi.org/10.11646/zootaxa.3946.1.3>

<http://zoobank.org/urn:lsid:zoobank.org:pub:E7630BC6-637C-49E9-9C6D-E9C26DFA4AA0>

## Two new species of *Amazophrynell* (Amphibia: Anura: Bufonidae) from Loreto, Peru

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### Abstract

*Amazophrynell* is a taxonomically poorly known bufonid genus with a pan-Amazonian distribution. A large part of this ambiguity comes from taxonomic uncertainties regarding the type species *A. minuta*. In this study we compare morphological and molecular data of topotypic specimens of *A. minuta* with all other nominal congeneric species. Based on these comparisons, we describe two new species. The first species, *A. amazonicola* sp. nov., differs from other recognized congeners by having a tip of snout with a small triangular protrusion (in dorsal and lateral view), spiculated body and basal webbing on fingers I and II. The second species, *A. matses* sp. nov., differs from congeners by the smallest snout to vent length of the genus, edges of nasal protrusion dilated and elliptical shape palmar tubercles. The two species are allopatric, where the first species is known to be associated with white sand forests (=campinaranas), while the second inhabits upland (=terra firme) forests. Both species are diagnosable by a series of substitutions in the 16S rDNA, and both species are highly divergent from their sister taxa (*p*-distances range from 7–14%).

**Key words:** Amazon basin, Riverine barriers, Biodiversity, Speciation

### Resumo

*Amazophrynell* é um gênero da família Bufonidae taxonomicamente pouco conhecido com uma distribuição pan-amazônica. Uma grande parte dessa ambiguidade vem de incertezas taxonômicas em relação com a espécie tipo *A. minuta*. Neste estudo, compararamos os dados morfológicos e moleculares de amostras topotípicas de *A. minuta* com todas as outras espécies nominais. Com base nessas comparações, descrevemos duas novas espécies. A primeira espécie, *A. amazonicola* sp. nov., difere dos outros congêneres reconhecidos por ter a ponta do focinho com uma pequena saliência triangular (vista dorsal e lateral), o corpo espiculado e membranas basais nos dedos I e II. A segunda espécie, *A. matses* sp. nov., difere de seus congêneres por apresentar o menor tamanho focinho-cloaca do gênero, bordas nasais com protuberâncias e tubérculos palmares de formas elípticas. As duas espécies são alopátricas, a primeira espécie é conhecida por estar associada com florestas de areia branca (= Campinaranas), enquanto que a segunda habita florestas de terras altas (= terra firme). Ambas as espécies são diagnosticáveis por uma série de substituições na região 16S do DNAr, e são altamente divergentes da sua espécie irmã (*p*-distâncias não corrigidas 7–14%).

### Resumen

*Amazophrynell* es un género de la familia Bufonidae taxonómicamente poco conocido con una distribución pan-amazónica. Una gran parte de esta ambigüedad proviene de incertidumbres taxonómicas en relación con la especie tipo

We are grateful to Mario Nunes for his assistance with laboratory work, and to Addison Wynn, Jeremy Jacobs and staff of USNM for their hospitality and support at the Smithsonian Institution. We thank the people of the community of Taracuá, especially Sr. Maximiliano Correia Menezes (Representative of the Amerindian village of Taracuá and FOIRN), Sr. Gabriel Correia Menezes and Sra. Vera Correia for their hospitality and logistic support in Taracuá, and the Federação de Organizações Indígenas do Rio Negro - FOIRN for permission to access indigenous land. RRR thanks Ian Pool Medina, Richard Curto, and Sally Ismodes for the support in fieldwork in Peru and Giussepe Gagliardi for photos.

## Literature cited

- Ávila, R.W., Carvalho, V.T., Gordo, M., Ribeiro, K. & Morais, D. (2012) A new species of *Amazophrynellula* (Anura: Bufonidae) from southern Amazonia. *Zootaxa*, 3484, 65–74.  
<http://dx.doi.org/10.2307/2419810>
- Baum, D.A. & Donoghue, M.J. (1995) Choosing among alternative “phylogenetic” species concepts. *Systematic Botany*, 20, 560–573.  
[http://dx.doi.org/10.1007/978-1-4615-6781-3\\_6](http://dx.doi.org/10.1007/978-1-4615-6781-3_6)
- Brown, J. & Twomey, E. (2009) Complicated histories: three new species of poison frogs of the genus *Ameerega* (Anura: Dendrobatidae) from north-central Peru. *Zootaxa*, 2049, 1–38.
- Cracraft, J. (1983) Species concepts and speciation analysis. *Current Ornithology*, 1, 159–187.  
[http://dx.doi.org/10.1007/978-1-4615-6781-3\\_6](http://dx.doi.org/10.1007/978-1-4615-6781-3_6)
- Crawford, A.J., Lips, K.R. & Bermingham, E. (2010) Epidemic disease decimates amphibian abundance, species diversity, and evolutionary history in the highlands of central Panama. *Proceedings of the National Academy of Sciences of the United States of America*, 107, 13777–13782.  
<http://dx.doi.org/10.1073/pnas.0914115107>
- Cruz, C.A.G. & Fusinatto, L.A. (2008) A new species of *Dendrophryniscus* Jiménes de la Espada, 1871 (Amphibia, Anura, Bufonidae) from the Atlantic Forest of Rio Grande do Sul, Brazil. *South American Journal of Herpetology*, 3, 22–26.  
[http://dx.doi.org/10.2994/1808-9798\(2008\)3\[22:ansodj\]2.0.co;2](http://dx.doi.org/10.2994/1808-9798(2008)3[22:ansodj]2.0.co;2)
- Davis, J.I. & Nixon, K.C. (1992) Populations, genetic variation, and the delimitation of phylogenetic species. *Systematic Biology*, 41, 421–435.  
<http://dx.doi.org/10.1093/sysbio/41.4.421>
- Darst, C.R. & Cannatella, D.C. (2004) Novel relationships among hyloid frogs inferred from 12S and 16S mitochondrial DNA sequences. *Molecular Phylogenetics and Evolution*, 31, 462–475.  
<http://dx.doi.org/10.1016/j.ympev.2003.09.003>
- de Queiroz, K. (2007) Species concepts and species delimitation. *Systematic Biology*, 56, 879–886.  
<http://dx.doi.org/10.1080/10635150701701083>
- Duellman, W.E. & Mendelson, J.R. (1995) Amphibians and reptiles from northern Departamento Loreto, Peru: Taxonomy and biogeography. *University of Kansas Science Bulletin*, 55, 329–376.
- Duellman, W.E. (1978) The biology of an equatorial herpetofauna in Amazonian Ecuador. *Miscellaneous Publications, University of Kansas Museum of Natural History*, 65, 1–352.
- Felsenstein, J. (1981) Evolutionary trees from DNA sequences: a maximum likelihood approach. *Journal of Molecular Evolution*, 17, 396–376.  
<http://dx.doi.org/10.1007/bf01734359>
- Felsenstein, J. (1981) Confidence limits on phylogenies: An approach using the bootstrap. *Evolution*, 39, 783–791.  
<http://dx.doi.org/10.2307/2408678>
- Fouquet, A., Gilles, A., Vences, M., Marty, C., Blanc, M. & Gemmell, N.J. (2007) Underestimation of species richness in Neotropical frogs revealed by mtDNA analyses. *PLoS ONE*, 2, e1109.  
<http://dx.doi.org/10.1371/journal.pone.0001109>
- Fouquet, A., Recoder, R., Teixeira Jr., M., Cassimiro, J., Amaro, R.C., Camacho, A., Damasceno, R., Carnaval, A.C., Moritz, C. & Rodrigues, M.T. (2012a) Molecular phylogeny and morphometric analyses reveal deep divergence between Amazonia and Atlantic Forest species of *Dendrophryniscus*. *Molecular Phylogenetics and Evolution*, 62, 823–838.  
<http://dx.doi.org/10.1016/j.ympev.2011.11.023>
- Fouquet, A., Recoder, R., Teixeira Jr., M., Cassimiro, J., Amaro, R.C., Camacho, A., Damasceno, R., Carnaval, A.C., Moritz, C. & Rodrigues, M.T. (2012b) *Amazonella* Fouquet et al. 2012 (Anura: Bufonidae) junior homonym of *Amazonella* Lundblad, 1931 (Acari: Unionicolidae): proposed replacement by *Amazophrynellula* nom. nov. *Zootaxa*, 3244, 68.  
<http://dx.doi.org/10.1111/j.1095-8312.2012.01871.x>
- Fouquet, A., Ledoux, J.B., Dubut, V., Noonan, B.P. & Scotti, I. (2012c) The interplay of dispersal limitation, rivers, and historical events shapes the genetic structure of an Amazonian frog. *Biological Journal of the Linnean Society*, 106, 356–337.  
<http://dx.doi.org/10.1111/j.1095-8312.2012.01871.x>
- Fouquet, A., Noonan, B.P., Rodrigues, M.T., Pech, N., Gilles, A. & Gemmell, N.J. (2012d) Multiple quaternary refugia in the

- eastern Guiana Shield revealed by comparative phylogeography of 12 frog species. *Systematic Biology*, 61, 1–28.  
<http://dx.doi.org/10.1093/sysbio/syr130>
- Frost, D.R. (2014) *Amphibian Species of the World: an Online Reference*. Version 5.4 (9 January, 2013) American Museum of Natural History, New York, USA. Available from: <http://research.amnh.org/herpetology/amphibia/> (accessed 10 January 2015)
- Gordo, M. (2006) In: Vriesendorp, C., Pitman, N., Rojas, M., Pawlak, B.A., Rivera, C., Calixto M., Vela C.P. & Fasabi, R. (Eds.), 2006. *Perú: Matsés. Rapid Biological Inventories Report 16*. The Field Museum, Chicago, Illinois, 336 pp.
- Izecksohn, E. (1993) Nova espécie de *Dendrophryniscus* da região amazônica (Amphibia, Anura, Bufonidae). *Revista Brasileira de Zoologia*, 10, 407–412.  
<http://dx.doi.org/10.1590/s0101-81751993000300006>
- Jobb, G. (2008) TREEFINDER version of March 2011. Distributed by the author, Munich, Germany. Available from: <http://www.treefinder.de/> (accessed 27 May 2013)
- Kok, P.J. & Kalamandeen, M. (2008) *Introduction to the taxonomy of the amphibians of Kaieteur National Park, Guyana*. Abc Taxa Volumen 5, 288 pp.
- Lima, A.P., Magnusson, W.E., Menin, M., Erdtmann, L.K., Rodrigues, D.J., Keller, C. & Hödl, W. (2006) *Guia de sapos da Reserva Adolpho Ducke, Amazônia Central – Guide to the frogs of Reserva Adolpho Ducke, Central Amazonian*. Áttema Design Editorial, Manaus, 168 pp.
- Magnusson, W.E. & Hero, J.H. (1991) Predation and the evolution of complex oviposition behaviour in Amazon rainforest frogs. *Oecologia*, 86, 310–318.  
<http://dx.doi.org/10.1007/bf00317595>
- Melin, D.E. (1941) Contributions to the knowledge of the Amphibia of South America. *Göteborgs Kungl. Vetenskaps- och Vitterhets-samhällses. Handlingar. Serien B, Matematiska och Naturvetenskapliga Skrifter*, 1, 1–71.
- Padial, J.M. & De la Riva, I. (2009) Integrative taxonomy reveals cryptic Amazonian species of *Pristimantis* (Anura: Strabomantidae). *Zoological Journal of the Linnean Society*, 155, 97–122.
- Padial, J.M., Miralles, A., De la Riva, I. & Vences, M. (2010) The integrative future of taxonomy. *Frontiers in Zoology*, 7, 16.  
<http://dx.doi.org/10.1186/1742-9994-7-16>
- Padial, J.M., Chaparro, J.C., Castro Viejo-Fisher, S., Guyasamin, J., Lehr, E., Delgado, A., Vaira, M., Texeira, J.R., Aguayo, R. & De la Riva, I. (2012) A revision of species diversity in the Neotropical genus *Oreobates* (Anura: Strabomantidae), with the description of three new species from the Amazonian slopes of the Andes. *American Museum Novitates*, 3755, 1–55.  
<http://dx.doi.org/10.1206/3752.2>
- Posada, D. (2006) ModelTest Server: a web-based tool for the statistical selection of models of nucleotide substitution online. *Nucleic Acids Research*, 1, 34.  
<http://dx.doi.org/10.1093/nar/gkl042>
- Pramuk, J.B., Robertson, B., Sites, J.W. & Noonan, B.P. (2008) Around the world in 10 million years: biogeography of the nearly cosmopolitan true toads (Anura: Bufonidae). *Global Ecology and Biogeography*, 17, 72–83.  
<http://dx.doi.org/10.1111/j.1466-8238.2007.00348.x>
- Rach, J., DeSalle, R., Sarkar, I.N., Schierwater, B. & Hadrys, H. (2008) Character-based DNA barcoding allows discriminations of genera, species and populations in Odonata. *Proceedings of the Royal Society of London*, 275, 237–247.  
<http://dx.doi.org/10.1098/rspb.2007.1290>
- Rodríguez, L.O. & Duellman, W.E. (1994) *Guide to the Frogs of the Iquitos Region. Amazonian Peru*. University of Kansas Natural History Museum, special Publications, 22, 1–80.
- Rojas, R.R., Carvalho, V.T., Gordo, M., Ávila, R.W., Farias, I.P. & Hrbek, T. (2014) A new species of *Amazophrynellia* (Anura: Bufonidae) from the southwestern part of the Brazilian Guiana Shield. *Zootaxa*, 3753 (1), 79–95.  
<http://dx.doi.org/10.11646/zootaxa.3753.1.7>
- Sambrook, J., Fritsch, E.F. & Maniatis, T. (1989) *Molecular Cloning: A Laboratory Manual, second edition*. Cold Spring Harbor Laboratory Press, Cold Springs Harbor, NY, 1626 pp.
- Simões, P.I., Lima, A.P., Magnusson, W.E., Hödl, W. & Amézquita, A. (2008) Acoustic and morphological differentiation in the frog *Allobates femoralis*: relationships with the upper Madeira River and other potential geological barriers. *Biotropica*, 40, 607–614.  
<http://dx.doi.org/10.1111/j.1744-7429.2008.00416.x>
- Tamura, K., Dudley, J., Nei, M. & Kumar, S. (2007) MEGA4: Molecular Evolutionary Genetics Analysis (MEGA) software version 4.0. *Molecular Biology and Evolution*, 24, 1596–1599.  
<http://dx.doi.org/10.1093/molbev/msm092>
- Vences, M., Meike, T., Van der Meijden, A., Chiari, Y. & Vieites, D. (2005) Comparative performance of the 16S rRNA gene in DNA barcoding of amphibians. *Frontiers in Zoology*, 2, 7–12.